

IN THE CLAIMS:

Please amend claims 3, 6, 11 - 12, 18-19, 22 - 30, 32 - 39, 41 - 42 and 46 as follows:

*a2*  
3. The influenza virus of claim 1, wherein the at least one distinguishing amino acid residue to be replaced is located within the PB1 segment of the virus.

*a3*  
6. The influenza virus according to claim 1, wherein the modified RNA-polymerase is capable of recognition of segments with modified vRNA promoter sequences resulting in an enhanced rate of transcription and/or replication relative to said wild-type human influenza virus RNA-polymerase.

*Sub A*  
11. The influenza virus according to claim 7, wherein the 5' terminal nucleotide sequence comprises the modifications U3A and A8U resulting in a 5'-terminal sequence of 5'-AGAAGAAUCAAGG.

12. The influenza virus which is suitable for high yielding expression of one or more foreign recombinant or altered viral proteins, preferably said influenza virus contains

*a4*  
(i) one or more segment(s) with a foreign recombinant or altered viral gene sequence in addition to the RNA segments of the normal viral genome (additional segment) or partially replacing them (replacing segment), whereby the additional segment(s) and replacing segment(s) comprise the foreign or altered gene encoding the protein to be expressed in monocistronic arrangement and have modified vRNA promoter sequences as defined in claim 7; and/or

94

(ii) one or more bicistronic vRNA segment(s), preferably in ambisense or in tandem arrangement, whereby the bicistronic vRNA segment(s) has/have foreign gene(s) encoding the protein(s) to be expressed and being in covalent linkage with one of the authentic viral genes, preferably the neuraminidase gene, and has/have modified vRNA promoter sequences as defined in claim 7.

18. The influenza virus of claim 16, wherein in the ambisense RNA molecule said foreign recombinant gene is covalently bound to one of the viral genes, while the original vRNA segment coding for the same gene is deleted from the recombinant virus by way of specific ribozyme cleavage or is left out from the set of RNA-polymerase I promoted vRNA synthesizing plasmids, able to result in infectious viruses.

95

19. The influenza virus according to claim 16, wherein one or more of the standard viral RNA segments, differing from said at least one ambisense RNA segment, comprises a vRNA encoding a foreign gene, preferably one or more of the regular viral RNA segments has (have) been exchanged for a vRNA encoding a foreign gene, preferably one or both of the standard glycoproteins hemagglutinin and neuraminidase have been exchanged into foreign glycoprotein(s) or into fusion glycoproteins consisting of an anchor segment derived from hemagglutinin and an ectodomain obtained from the foreign source, viral or cellular, or in which such recombinant glycoprotein has been inserted as a third molecular species in addition to the remaining standard components.

22. The influenza virus of claim 20, wherein at least one of the regular viral RNA segments is replaced by a tandem RNA segment, preferably the replaced regular viral RNA segment is selected from the neuraminidase segment, hemagglutinin segment and NS segment.

96

23. The influenza virus according to claim 20, wherein the splice donor and splice acceptor signals are selected from sequences as present in influenza WSN segment 7 and 8 or other

partially effective splice reacting substrates, preferably the splice donor and splice acceptor signals are selected from sequences as present in influenza WSN segment 7.

24. The influenza virus according to claims 20, wherein one or more of the regular viral RNA segments, differing from said at least one tandem RNA segment, comprises a vRNA encoding a foreign gene which may or may not be in covalent connection to one of the viral genes, and preferably one or more of the regular viral RNA segments has (have) been deleted and replaced by a tandem vRNA encoding in addition a foreign gene.

25. The influenza virus according to claim 20, in which the foreign gene(s) in the tandem RNA segment

- (i) code for proteins and/or glycoproteins which are secreted from cells infected with the recombinant virus;
- (ii) code for proteins or artificial polypeptides designed to support an efficient HLA-restricted presentation of inherent epitopes at the surface of infected cells, for stimulation of a B cell and/or T cell response;
- (iii) is a nucleotide sequence causing viral attenuation, preferably the foreign gene is coding for part of the viral neuraminidase gene in inverted, i.e. sense orientation, with or without an inserted ribozyme sequence,

preferably the tandem segment part of the neuraminidase gene in sense orientation is attached to the hemagglutinin vRNA segment, and optionally another gene or reporter gene is encoded in a second tandem vRNA, preferably in conjunction with NS2.

26. The influenza virus according to claim 16 which is suitable for the expression of non-influenza genes or synthetic genes, or gene-inhibitory sequences such as, but not limited to, antisense genes or ribozymes, whereby

- (i) the non-influenza genes are covalently linked to one of the viral genes,
- (ii) the non-influenza gene constitutes a membrane glycoprotein consisting of a fusion of the viral HA transmembrane and cytoplasmic regions with the foreign ectodomain sequence.

27. A non-avian, non-human influenza virus, preferably an equine or a porcine influenza virus comprising an RNA-sequence encoding a modified RNA-polymerase which differs from the wild-type RNA-polymerase of said non-avian, non-human influenza virus in that at least one of the amino acid residue(s) distinguishing the wild-type RNA-polymerase of said non-avian, non-human influenza virus from FPV Bratislava RNA-polymerase has been replaced with the corresponding amino acid residue(s) as present in FPV Bratislava RNA-polymerase, preferably said influenza virus is as defined in claim 2.

28. A process for preparing the influenza virus of claim 1 which comprises replacing the RNA-sequence encoding the wild-type RNA-polymerase of said influenza virus with an RNA-sequence encoding the modified RNA-polymerase.

29. The process of claim 28, which is suitable for preparing PB1-chimeric viruses as well as recombinant viruses, said viruses being generated via cotransfection of up to eight cDNA plasmids containing the viral cDNAs, or chimeric (segment 2: PB1) and bicistronic recombinant (segment 6: NA/foreign gene) cDNA sequences instead, in such a way that they are transcribed *in vivo* by both RNA-polymerase I and RNA-polymerase II and jointly give rise to progeny viruses according to the plasmid insert design.

30. A pharmaceutical composition comprising the influenza virus according to claim 1.

32. Method of using the influenza virus according to claim 1 for preparing an agent

- (i) for gene transfer into cells, preferably into mammalian cells, more preferably into human cells, by viral infection;
- (ii) for gene transfer into antigen-presenting cells and the use of the obtained product for *ex vivo* immunotherapy;
- (iii) for *in vivo* somatic gene therapy;
- (iv) for *in vivo* vaccination, including therapeutic and prophylactic vaccination;
- (v) for eliciting an immune response, including the induction of a T-cell response;
- (vi) for treating a growing tumor or a chronic infectious disease.

33. A method for

- (i) gene transfer into cells, preferably into mammalian cells, more preferably into human cells, by viral infection;
- (ii) gene transfer into antigen-presenting cells, and the use of the obtained product for *ex vivo* immunotherapy;
- (iii) *in vivo* somatic gene therapy;
- (iv) *in vivo* vaccination, including therapeutic and prophylactic vaccination;
- (v) eliciting an immune response, including the induction of a T-cell response, preferably a CD4+ T-cell response, a CD8 T-cell response or both, or the induction of an antibody response;
- (vi) treating a growing tumor or a chronic infectious disease;
- (vii) preparing a vaccine;
- (viii) preventing and/or treating influenza;

which comprises contacting the cells, the antigen-presenting cells, the person or the patient in need for vaccination, for influenza treatment or for somatic gene therapy, or cell cultures with the influenza virus according to claim 1.

34. A method for the production of proteins or glycoproteins which comprises utilizing the influenza virus according to claim 1 as expression vector, preferably the production method is performed in cell culture cells or in fertilized chicken eggs.

35. Method of using the influenza virus according to claim 1 for preparing agents  
(i) for transfer and expression of foreign genes into cells infected by such viruses, or  
(ii) for transfer and expression of RNA molecules into cells infected by such viruses, preferably the RNA molecules to be expressed are antisense sequences or double-strand sequences relative to the target cell cellular mRNA molecules, and/or the agent is suitable for sequence-specific gene silencing, preferably by antisense RNA or RNA interference mechanisms such as ribozyme cleavages of target RNAs.

36. A method for transfer and expression of foreign genes into cells, and for transfer and expression of RNA molecules into cells, which method comprises infecting the cells with the influenza virus according to claim 1.

37. Method of using the influenza virus according to claim 1 for preparing agents for immunotherapy, preferably for autologous immunotherapy.

38. A method for an immunotherapy which comprises *ex vivo* infection of immune cells, preferably dendritic cells, with the influenza virus according to claim 1, and introduction of the transduced cells into the patient.

*a7*  
39. A method to elicit an immune response directed against an antigen, comprising the steps of introducing the influenza virus as defined in claim 1, into a cell or administering it to a mammal, wherein said influenza virus contains at least one foreign gene encoding the antigen.

*a8*  
41. The method of claim 39, wherein the polynucleotide sequence

- (i) is derivable from a cDNA library isolated from tumor cells, or testis cells, or virus-infected cells, or microbially infected cells, or cell-lines,
- (ii) is a fusion protein consisting of epitopes derived from one or more T-cell specific epitope sequences as present in viral or other pathogens, or in tumor associated antigens.

*a9*  
42. A vaccine for therapeutic or prophylactic purposes which is a human influenza virus vaccine comprising a human influenza virus as defined in claim 1, preferably said human influenza virus encodes the antigen for a membrane protein and in addition contains the membrane protein in the viral envelope.

*a10*  
46. A method to identify a polynucleotide sequence encoding at least one HLA-restricted epitope comprising the steps of

- (a) preparing a gene bank or a cDNA bank from the cell or the microorganism to be tested;
- (b) incorporating the cDNA or the DNA of the gene bank into the genome of the influenza virus as defined in claim 1 to yield recombinant virus particles,
- (c) infecting immortalized autologous cells, which are capable of expression of HLA-class I molecules and/or HLA-class II molecules on their surface, with the recombinant virus particles obtained in step (b),

- (d) expressing the proteins encoded by said cDNA or said DNA of the gene bank in the autologous cells and presenting the fragments of the proteins produced by the autologous cells or the cell surface in connection with HLA molecules;
- (e) co-cultivating T-cells with the autologous cells; and
- (f) stimulating the T-cells by such autologous cells which present antigens on their surface, whereby said antigens are recognized by the T-cells.

Please add the following new claims:

48. A vaccine for therapeutic or prophylactic purposes which is a non-human influenza virus vaccine, preferably an equine or porcine influenza virus vaccine, comprising a virus as defined in claim 27.

49. A pharmaceutical composition comprising the influenza virus according to claim 27.

50. A method for transfer and expression of foreign genes into cells, and for transfer and expression of RNA molecules into cells, which method comprises infecting the cells with the influenza virus according to claim 27.

51. A method for an immunotherapy which comprises *ex vivo* infection of immune cells, preferably dendritic cells, with the influenza virus according to claim 27, and introduction of the transduced cells into the patient.

52. A method to elicit an immune response directed against an antigen, comprising the steps of introducing the influenza virus as defined in claim 27, into a cell or administering it to a mammal, wherein said influenza virus contains at least one foreign gene encoding the antigen.